Q2(R1) Validation of Analytical Procedures: Text and Methodology

Guidance for Industry

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
Center for Biologics Evaluation and Research (CBER)

November 2005[1]
ICH

[1] This guidance, *Q2(R1) Validation of Analytical Procedures: Text and Methodology* (Q2(R1)), was initially published as two separate guidances, *Q2A Text on Validation of Analytical Procedures* (March 1995)(Q2A) and *Q2B Validation of Analytical Procedures: Methodology* (May 1997)(Q2B). In November 2005, the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) incorporated Q2B on methodology with the parent document Q2A and retitled the combined document Q2(R1). In October 2021, FDA likewise incorporated Q2B with the parent document Q2A and retitled the combined document Q2(R1). This guidance consists of the previously published FDA guidances Q2A and Q2B. It is the same, in substance, as those two guidances, and it is the same, in substance, as the November 2005 ICH Q2(R1) guideline.

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Q2(R1)
Document History

Document Codification	Document Name	Date
Q2A	Q2A Text on Validation of Analytical Procedures	March 1995
Q2B	Q2B Validation of Analytical Procedures: Methodology	May 1997
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[3] As noted above, in October 2021, FDA incorporated *Q2B Validation of Analytical Procedures: Methodology* (May 1997)(Q2B) on methodology with the parent document *Q2A Text on Validation of Analytical Procedures* (March 1995)(Q2A) and retitled the combined document *Q2(R1) Validation of Analytical Procedures: Text and Methodology* (Q2(R1). This guidance consists of the previously published FDA guidances, Q2A and Q2B. It is the same, in substance, as those two guidances, and it is the same, in substance, as the November 2005 ICH Q2(R1) guideline.

FOREWORD

The International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) has the mission of achieving greater regulatory harmonization worldwide to ensure that safe, effective, and high-quality medicines are developed, registered, and maintained in the most resource-efficient manner. By harmonizing the regulatory expectations in regions around the world, ICH guidelines have substantially reduced duplicative clinical studies, prevented unnecessary animal studies, standardized safety reporting and marketing application submissions, and contributed to many other improvements in the quality of global drug development and manufacturing and the products available to patients.

ICH is a consensus-driven process that involves technical experts from regulatory authorities and industry parties in detailed technical and science-based harmonization work that results in the development of ICH guidelines. The commitment to consistent adoption of these consensus-based guidelines by regulators around the globe is critical to realizing the benefits of safe, effective, and high-quality medicines for patients as well as for industry. As a Founding Regulatory Member of ICH, the Food and Drug Administration (FDA) plays a major role in the development of each of the ICH guidelines, which FDA then adopts and issues as guidance to industry.

NOTE

This guidance consists of two previously published FDA guidances that were developed and finalized before the implementation of FDA's good guidance practices regulation (21 CFR 10.115).[4]

[4] As noted above, in October 2021, FDA incorporated *Q2B Validation of Analytical Procedures: Methodology* (May 1997)(Q2B) on methodology with the parent document *Q2A Text on Validation of Analytical Procedures* (March 1995)(Q2A) and retitled the combined document *Q2(R1) Validation of Analytical Procedures: Text and Methodology* (Q2(R1). This guidance consists of the previously published FDA guidances, Q2A and Q2B, without any substantive changes.

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Contains Nonbinding Recommendations

Q2(R1) Validation of Analytical Procedures: Text and Methodology Guidance for Industry

This guidance represents the current thinking of the Food and Drug Administration (FDA or Agency) on this topic. It does not establish any rights for any person and is not binding on FDA or the public. You can use an alternative approach if it satisfies the requirements of the applicable statutes and regulations. To discuss an alternative approach, contact the FDA office responsible for this guidance as listed on the title page.

PART I[5]

TEXT ON VALIDATION OF ANALYTICAL PROCEDURES

I. INTRODUCTION

This document presents a discussion of the characteristics for consideration during the validation of the analytical procedures included as part of registration applications submitted within the European Union, Japan, and United States. This document does not necessarily seek to cover the testing that may be required for registration in, or export to, other areas of the world. Furthermore, this text presentation serves as a collection of terms, and their definitions, and is not intended to provide direction on how to accomplish validation. These terms and definitions are meant to bridge the differences that often exist between various compendia and regulators of the European Union, Japan, and United States.

The objective of validation of an analytical procedure is to demonstrate that it is suitable for its intended purpose. A tabular summation of the characteristics applicable to identification, control of impurities and assay procedures is included. Other analytical procedures may be considered in future additions to this document.

In general, FDA's guidance documents do not establish legally enforceable responsibilities. Instead, guidances describe the Agency's current thinking on a topic and should be viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word *should* in Agency guidances means that something is suggested or recommended, but not required.

[5] Part I of this guidance was developed within the Expert Working Group (Quality) of the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) and has been subject to consultation by the regulatory parties, in accordance with the ICH process. This document has been endorsed by the ICH Steering Committee at *Step 4* of the ICH process, October 27, 1994. At *Step 4* of the process, the final draft is recommended for adoption to the regulatory bodies of the ICH regions. This guidance is applicable to drug and biological products.

II. TYPES OF ANALYTICAL PROCEDURES TO BE VALIDATED

The discussion of the validation of analytical procedures is directed to the four most common types of analytical procedures:

- Identification tests.
- Quantitative tests for impurities' content.
- Limit tests for the control of impurities.
- Quantitative tests of the active moiety in samples of drug substance or drug product or other selected component(s) in the drug product.

Although there are many other analytical procedures, such as dissolution testing for drug products or particle size determination for drug substance, these have not been addressed in the initial text on validation of analytical procedures. Validation of these additional analytical procedures is equally important to those listed herein and may be addressed in subsequent documents.

A brief description of the types of tests considered in this document is provided below.

- Identification tests are intended to ensure the identity of an analyte in a sample. This is normally achieved by comparison of a property of the sample (e.g., spectrum, chromatographic behavior, chemical reactivity, etc.) to that of a reference standard.
- Testing for impurities can be either a quantitative test or a limit test for the impurity in a sample. Either test is intended to accurately reflect the purity characteristics of the sample. Different validation characteristics are required for a quantitative test than for a limit test.
- Assay procedures are intended to measure the analyte present in a given sample. In the context of this document, the assay represents a quantitative measurement of the major component(s) in the drug substance. For the drug product, similar validation characteristics also apply when assaying for the active or other selected component(s). The same validation characteristics may also apply to assays associated with other analytical procedures (e.g., dissolution).

The objective of the analytical procedure should be clearly understood since this will govern the validation characteristics which need to be evaluated. Typical validation characteristics which should be considered are listed below:

- Accuracy
- Precision
  - Repeatability
  - Intermediate Precision
- Specificity

- Detection Limit
- Quantitation Limit
- Linearity
- Range

Each of these validation characteristics is defined in the attached Glossary. The table lists those validation characteristics regarded as the most important for the validation of different types of analytical procedures. This list should be considered typical for the analytical procedures cited but occasional exceptions should be dealt with on a case-by-case basis. It should be noted that robustness is not listed in the table but should be considered at an appropriate stage in the development of the analytical procedure.

Furthermore, revalidation may be necessary in the following circumstances:

- changes in the synthesis of the drug substance;
- changes in the composition of the finished product;
- changes in the analytical procedure.

The degree of revalidation required depends on the nature of the changes. Certain other changes may require validation as well.

TABLE

TYPE OF ANALYTICAL PROCEDURE; CHARACTERISTICS	IDENTIFICATION	TESTING FOR IMPURITIES		ASSAY; dissolution (measurement only); content/potency
		Quantitation	Limit	
Accuracy	-	+	-	+
Precision				
Repeatability	-	+	-	+
Interm. Precision	-	+ [1]	-	+ [1]
Specificity [2]	+	+	+	+
Detection Limit	-	- [3]	+	-
Quantitation Limit	-	+	-	-
Linearity	-	+	-	+
Range	-	+	-	+

NOTE: - signifies that this characteristic is not normally evaluated; + signifies that this characteristic is normally evaluated

[1] in cases where reproducibility (see Glossary) has been performed, intermediate precision is not needed

[2] lack of specificity of one analytical procedure could be compensated by other supporting analytical procedure(s)

[3] may be needed in some cases

APPENDIX A. GLOSSARY

1. ANALYTICAL PROCEDURE

The analytical procedure refers to the way of performing the analysis. It should describe in detail the steps necessary to perform each analytical test. This may include but is not limited to: the sample, the reference standard and the reagents preparations, use of the apparatus, generation of the calibration curve, use of the formulae for the calculation, etc.

2. SPECIFICITY

Specificity is the ability to assess unequivocally the analyte in the presence of components which may be expected to be present. Typically these might include impurities, degradants, matrix, etc.

Lack of specificity of an individual analytical procedure may be compensated by other supporting analytical procedure(s).

This definition has the following implications:

Identification: to ensure the identity of an analyte.

Purity Tests: to ensure that all the analytical procedures performed allow an accurate statement of the content of impurities of an analyte, i.e. related substances test, heavy metals, residual solvents content, etc.

Assay (content or potency):
to provide an exact result which allows an accurate statement on the content or potency of the analyte in a sample.

3. ACCURACY

The accuracy of an analytical procedure expresses the closeness of agreement between the value which is accepted either as a conventional true value or an accepted reference value and the value found.

This is sometimes termed trueness.

4. PRECISION

The precision of an analytical procedure expresses the closeness of agreement (degree of scatter) between a series of measurements obtained from multiple sampling of the same homogeneous sample under the prescribed conditions. Precision may be considered at three levels: repeatability, intermediate precision and reproducibility.

Precision should be investigated using homogeneous, authentic samples. However, if it is not possible to obtain a homogeneous sample it may be investigated using artificially prepared samples or a sample solution.

The precision of an analytical procedure is usually expressed as the variance, standard deviation or coefficient of variation of a series of measurements.

a. Repeatability

Repeatability expresses the precision under the same operating conditions over a short interval of time. Repeatability is also termed intra-assay precision.

b. Intermediate precision

Intermediate precision expresses within-laboratories variations: different days, different analysts, different equipment, etc.

c. Reproducibility

Reproducibility expresses the precision between laboratories (collaborative studies, usually applied to standardization of methodology).

5. DETECTION LIMIT

The detection limit of an individual analytical procedure is the lowest amount of analyte in a sample which can be detected but not necessarily quantitated as an exact value.

6. QUANTITATION LIMIT

The quantitation limit of an individual analytical procedure is the lowest amount of analyte in a sample which can be quantitatively determined with suitable precision and accuracy. The quantitation limit is a parameter of quantitative assays for low levels of compounds in sample matrices, and is used particularly for the determination of impurities and/or degradation products.

7. LINEARITY

The linearity of an analytical procedure is its ability (within a given range) to obtain test results which are directly proportional to the concentration (amount) of analyte in the sample.

8. RANGE

The range of an analytical procedure is the interval between the upper and lower concentration (amounts) of analyte in the sample (including these concentrations) for which it has been demonstrated that the analytical procedure has a suitable level of precision, accuracy and linearity.

9. ROBUSTNESS

The robustness of an analytical procedure is a measure of its capacity to remain unaffected by small, but deliberate variations in method parameters and provides an indication of its reliability during normal usage.

PART II[6]

VALIDATION OF ANALYTICAL PROCEDURES: METHODOLOGY

I. INTRODUCTION

This document is complementary to the parent document, which presents a discussion of the characteristics that should be considered during the validation of analytical procedures. Its purpose is to provide some guidance and recommendations on how to consider the various validation characteristics for each analytical procedure. In some cases (for example, demonstration of specificity), the overall capabilities of a number of analytical procedures in combination may be investigated in order to ensure the quality of the drug substance or drug product. In addition, the document provides an indication of the data that should be presented in a new drug application.

All relevant data collected during validation and formulae used for calculating validation characteristics should be submitted and discussed as appropriate.

Approaches other than those set forth in this guidance may be applicable and acceptable. It is the responsibility of the applicant to choose the validation procedure and protocol most suitable for their product. However, it is important to remember that the main objective of validation of an analytical procedure is to demonstrate that the procedure is suitable for its intended purpose. Due to their complex nature, analytical procedures for biological and biotechnological products in some cases may be approached differently than in this document.

Well-characterized reference materials, with documented purity, should be used throughout the validation study. The degree of purity necessary depends on the intended use.

In accordance with the parent document, and for the sake of clarity, this document considers the various validation characteristics in distinct sections. The arrangement of these sections reflects the process by which an analytical procedure may be developed and evaluated.

In practice, it is usually possible to design the experimental work such that the appropriate validation characteristics can be considered simultaneously to provide a sound, overall knowledge of the capabilities of the analytical procedure, for instance: Specificity, linearity, range, accuracy, and precision.

[6] Part II of this guidance was developed within the Expert Working Group (Quality) of the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) and has been subject to consultation by the regulatory parties, in accordance with the ICH process. This document has been endorsed by the ICH Steering Committee at *Step 4* of the ICH process, November 1996. At *Step 4* of the process, the final draft is recommended for adoption to the regulatory bodies of the ICH regions. This guidance is applicable to drug and biological products.

II. SPECIFICITY (1)

An investigation of specificity should be conducted during the validation of identification tests, the determination of impurities, and the assay. The procedures used to demonstrate specificity will depend on the intended objective of the analytical procedure.

It is not always possible to demonstrate that an analytical procedure is specific for a particular analyte (complete discrimination). In this case, a combination of two or more analytical procedures is recommended to achieve the necessary level of discrimination.

A. Identification (1.1)

Suitable identification tests should be able to discriminate between compounds of closely related structures which are likely to be present. The discrimination of a procedure may be confirmed by obtaining positive results (perhaps by comparison with a known reference material) from samples containing the analyte, coupled with negative results from samples which do not contain the analyte. In addition, the identification test may be applied to materials structurally similar to or closely related to the analyte to confirm that a positive response is not obtained. The choice of such potentially interfering materials should be based on sensible scientific judgment with a consideration of the interferences that could occur.

B. Assay and Impurity Test(s) (1.2)

For chromatographic procedures, representative chromatograms should be used to demonstrate specificity, and individual components should be appropriately labeled. Similar considerations should be given to other separation techniques.

Critical separations in chromatography should be investigated at an appropriate level. For critical separations, specificity can be demonstrated by the resolution of the two components which elute closest to each other.

In cases where a nonspecific assay is used, other supporting analytical procedures should be used to demonstrate overall specificity. For example, where a titration is adopted to assay the drug substance for release, the combination of the assay and a suitable test for impurities can be used.

The approach is similar for both assay and impurity tests:

1. Impurities are available (1.2.1)

For the assay, this should involve demonstration of the discrimination of the analyte in the presence of impurities and/or excipients; practically, this can be done by spiking pure substances (drug substance or drug product) with appropriate levels of impurities and/or excipients and demonstrating that the assay result is unaffected by the presence of these materials (by comparison with the assay result obtained on unspiked samples). For the impurity test, the discrimination may be established by spiking drug substance or drug product with appropriate

levels of impurities and demonstrating the separation of these impurities individually and/or from other components in the sample matrix.

2. Impurities are not available (1.2.2)

If impurity or degradation product standards are unavailable, specificity may be demonstrated by comparing the test results of samples containing impurities or degradation products to a second well-characterized procedure, e.g., pharmacopoeial method or other validated analytical procedure (independent procedure). As appropriate, this should include samples stored under relevant stress conditions: Light, heat, humidity, acid/base hydrolysis, and oxidation.

- For the assay, the two results should be compared.

- For the impurity tests, the impurity profiles should be compared.

Peak purity tests may be useful to show that the analyte chromatographic peak is not attributable to more than one component (e.g., diode array, mass spectrometry).

III. LINEARITY (2)

A linear relationship should be evaluated across the range (see section 3) of the analytical procedure. It may be demonstrated directly on the drug substance (by dilution of a standard stock solution) and/or separate weighings of synthetic mixtures of the drug product components, using the proposed procedure. The latter aspect can be studied during investigation of the range.

Linearity should be evaluated by visual inspection of a plot of signals as a function of analyte concentration or content. If there is a linear relationship, test results should be evaluated by appropriate statistical methods, for example, by calculation of a regression line by the method of least squares. In some cases, to obtain linearity between assays and sample concentrations, the test data may have to be subjected to a mathematical transformation prior to the regression analysis. Data from the regression line itself may be helpful to provide mathematical estimates of the degree of linearity.

The correlation coefficient, y-intercept, slope of the regression line, and residual sum of squares should be submitted. A plot of the data should be included. In addition, an analysis of the deviation of the actual data points from the regression line may also be helpful for evaluating linearity.

Some analytical procedures, such as immunoassays, do not demonstrate linearity after any transformation. In this case, the analytical response should be described by an appropriate function of the concentration (amount) of an analyte in a sample.

For the establishment of linearity, a minimum of five concentrations is recommended. Other approaches should be justified.

IV. RANGE (3)

The specified range is normally derived from linearity studies and depends on the intended application of the procedure. It is established by confirming that the analytical procedure provides an acceptable degree of linearity, accuracy, and precision when applied to samples containing amounts of analyte within or at the extremes of the specified range of the analytical procedure.

The following minimum specified ranges should be considered.

- For the assay of a drug substance or a finished (drug) product: Normally from 80 to 120 percent of the test concentration;

- For content uniformity: Covering a minimum of 70 to 130 percent of the test concentration, unless a wider, more appropriate range, based on the nature of the dosage form (e.g., metered dose inhalers), is justified;

- For dissolution testing: +/-20 percent over the specified range; e.g., if the specifications for a controlled released product cover a region from 20 percent, after 1 hour, up to 90 percent, after 24 hours, the validated range would be 0-110 percent of the label claim;

- For the determination of an impurity: From the reporting level of an impurity[7] to 120 percent of the specification;

- For impurities known to be unusually potent or to produce toxic or unexpected pharmacological effects, the detection/quantitation limit should be commensurate with the level at which the impurities must be controlled.

  Note: For validation of impurity test procedures carried out during development, it may be necessary to consider the range around a suggested (probable) limit;

- If assay and purity are performed together as one test and only a 100 percent standard is used, linearity should cover the range from the reporting level of the impurities[8] to 120 percent of the assay specification.

V. ACCURACY (4)

Accuracy should be established across the specified range of the analytical procedure.

A. Assay (4.1)

[7] See sections on "Reporting Impurity Content of Batches" of the corresponding guidance ICH Q3A *Impurities in New Drug Substances* (January 1996) and ICH Q3B guidance *Impurities in New Drug Products* (May 1997).
[8] See footnote 7.

1. Drug substance (4.1.1)

Several methods of determining accuracy are available.

(a) Application of an analytical procedure to an analyte of known purity (e.g., reference material);

(b) Comparison of the results of the proposed analytical procedure with those of a second well-characterized procedure, the accuracy of which is stated and/or defined (independent procedure, see section 1.2.);

(c) Accuracy may be inferred once precision, linearity, and specificity have been established.

2. Drug product (4.1.2)

Several methods for determining accuracy are available.

(a) Application of the analytical procedure to synthetic mixtures of the drug product components to which known quantities of the drug substance to be analyzed have been added;

(b) In cases where it is impossible to obtain samples of all drug product components, it may be acceptable either to add known quantities of the analyte to the drug product or to compare the results obtained from a second, well-characterized procedure, the accuracy of which is stated and/or defined (independent procedure, see section 1.2);

(c) Accuracy may be inferred once precision, linearity, and specificity have been established.

B. Impurities (Quantitation) (4.2)

Accuracy should be assessed on samples (drug substance/drug product) spiked with known amounts of impurities.

In cases where it is impossible to obtain samples of certain impurities and/or degradation products, it is considered acceptable to compare results obtained by an independent procedure (see section 1.2.). The response factor of the drug substance can be used.

It should be clear how the individual or total impurities are to be determined, e.g., weight/weight or area percent, in all cases with respect to the major analyte.

C. Recommended Data (4.3)

Accuracy should be assessed using a minimum of 9 determinations over a minimum of 3 concentration levels covering the specified range (e.g., 3 concentrations/3 replicates each of the total analytical procedure).

Accuracy should be reported as percent recovery by the assay of known added amount of analyte in the sample or as the difference between the mean and the accepted true value together with the confidence intervals.

VI. PRECISION (5)

Validation of tests for assay and for quantitative determination of impurities includes an investigation of precision.

A. Repeatability (5.1)

Repeatability should be assessed using:

(1) A minimum of 9 determinations covering the specified range for the procedure (e.g., 3 concentrations/3 replicates each); or

(2) A minimum of 6 determinations at 100 percent of the test concentration.

B. Intermediate Precision (5.2)

The extent to which intermediate precision should be established depends on the circumstances under which the procedure is intended to be used. The applicant should establish the effects of random events on the precision of the analytical procedure. Typical variations to be studied include days, analysts, equipment, etc. It is not necessary to study these effects individually. The use of an experimental design (matrix) is encouraged.

C. Reproducibility (5.3)

Reproducibility is assessed by means of an interlaboratory trial. Reproducibility should be considered in case of the standardization of an analytical procedure, for instance, for inclusion of procedures in pharmacopoeias. These data are not part of the marketing authorization dossier.

D. Recommended Data (5.4)

The standard deviation, relative standard deviation (coefficient of variation), and confidence interval should be reported for each type of precision investigated.

VII. DETECTION LIMIT (6)

Several approaches for determining the detection limit are possible, depending on whether the procedure is noninstrumental or instrumental. Approaches other than those listed below may be acceptable.

A. Based on Visual Evaluation (6.1)

Visual evaluation may be used for noninstrumental methods but may also be used with instrumental methods.

The detection limit is determined by the analysis of samples with known concentrations of analyte and by establishing the minimum level at which the analyte can be reliably detected.

B. Based on Signal-to-Noise (6.2)

This approach can only be applied to analytical procedures which exhibit baseline noise. Determination of the signal-to-noise ratio is performed by comparing measured signals from samples with known low concentrations of analyte with those of blank samples and establishing the minimum concentration at which the analyte can be reliably detected. A signal-to-noise ratio between 3 or 2:1 is generally considered acceptable for estimating the detection limit.

C. Based on the Standard Deviation of the Response and the Slope (6.3)

The detection limit (DL) may be expressed as:

$$DL = \frac{3.3\sigma}{S}$$

where σ = the standard deviation of the response
S = the slope of the calibration curve

The slope S may be estimated from the calibration curve of the analyte. The estimate of σ may be carried out in a variety of ways, for example:

1. Based on the standard deviation of the blank (6.3.1)

Measurement of the magnitude of analytical background response is performed by analyzing an appropriate number of blank samples and calculating the standard deviation of these responses.

2. Based on the calibration curve (6.3.2)

A specific calibration curve should be studied using samples containing an analyte in the range of DL. The residual standard deviation of a regression line or the standard deviation of y-intercepts of regression lines may be used as the standard deviation.

D. Recommended Data (6.4)

The detection limit and the method used for determining the detection limit should be presented. If DL is determined based on visual evaluation or based on signal-to-noise ratio, the presentation of the relevant chromatograms is considered acceptable for justification.

In cases where an estimated value for the detection limit is obtained by calculation or extrapolation, this estimate may subsequently be validated by the independent analysis of a suitable number of samples known to be near or prepared at the detection limit.

VIII. QUANTITATION LIMIT (7)

Several approaches for determining the quantitation limit are possible, depending on whether the procedure is noninstrumental or instrumental. Approaches other than those listed below may be acceptable.

A. Based on Visual Evaluation (7.1)

Visual evaluation may be used for noninstrumental methods but may also be used with instrumental methods.

The quantitation limit is generally determined by the analysis of samples with known concentrations of analyte and by establishing the minimum level at which the analyte can be quantified with acceptable accuracy and precision.

B. Based on Signal-to-Noise (7.2)

This approach can only be applied to analytical procedures that exhibit baseline noise. Determination of the signal-to-noise ratio is performed by comparing measured signals from samples with known low concentrations of analyte with those of blank samples and by establishing the minimum concentration at which the analyte can be reliably quantified. A typical signal-to-noise ratio is 10:1.

C. Based on the Standard Deviation of the Response and the Slope (7.3)

The quantitation limit (QL) may be expressed as:

$$QL = \frac{10\sigma}{S}$$

where σ = the standard deviation of responses
S = the slope of the calibration curve

The slope S may be estimated from the calibration curve of the analyte. The estimate of σ may be carried out in a variety of ways, for example:

1. Based on standard deviation of the blank (7.3.1)

Measurement of the magnitude of analytical background response is performed by analyzing an appropriate number of blank samples and calculating the standard deviation of these responses.

2. Based on the calibration curve (7.3.2.)

A specific calibration curve should be studied using samples containing an analyte in the range of QL. The residual standard deviation of a regression line or the standard deviation of y-intercepts of regression lines may be used as the standard deviation.

D. Recommended Data (7.4)

The quantitation limit and the method used for determining the quantitation limit should be presented.

The limit should be subsequently validated by the analysis of a suitable number of samples known to be near or prepared at the quantitation limit.

IX. ROBUSTNESS (8)

The evaluation of robustness should be considered during the development phase and depends on the type of procedure under study. It should show the reliability of an analysis with respect to deliberate variations in method parameters.

If measurements are susceptible to variations in analytical conditions, the analytical conditions should be suitably controlled or a precautionary statement should be included in the procedure. One consequence of the evaluation of robustness should be that a series of system suitability parameters (e.g., resolution test) is established to ensure that the validity of the analytical procedure is maintained whenever used.

Examples of typical variations are:

- Stability of analytical solutions
- Extraction time

In the case of liquid chromatography, examples of typical variations are:

- Influence of variations of pH in a mobile phase
- Influence of variations in mobile phase composition
- Different columns (different lots and/or suppliers)
- Temperature
- Flow rate

In the case of gas-chromatography, examples of typical variations are:

- Different columns (different lots and/or suppliers)
- Temperature
- Flow rate

X. SYSTEM SUITABILITY TESTING (9)

System suitability testing is an integral part of many analytical procedures. The tests are based on the concept that the equipment, electronics, analytical operations, and samples to be analyzed constitute an integral system that can be evaluated as such. System suitability test parameters to be established for a particular procedure depend on the type of procedure being validated. See pharmacopoeias for additional information.